Role of the Immune System(s) in Progressive MS

CMSC Annual Meeting
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Disclosures

I have participated as a speaker at meetings sponsored by, received consulting fees, and/or grant support from:

Biogen Idec, Diogenix, Genentech, Sanofi-Genzyme EMD/Merck Serono, GSK, Mitsubishi Pharma, Novartis, Receptos, Roche,, Teva Neuroscience

Investigational and off label agents may be discussed.
Concepts of inflammation and degeneration in MS:
[Inflammation throughout MS spectrum (lesion axon loss → microglial activation); divide into periph infilt vs CNS compartm]

Mechanisms:
Adaptive (T and B specificity; ? Target/ pediatric Axoglial?) vs Innate (bystander; chronic microglial activation – more later)

Direct inflamma damage vs. indirect (impacting degeneration – eg OPC failed repair, astrocyte support, of OPC, neurons eg NK; exacerbate metabolic stress/demand/mitochondria, channelopathies etc

Single vs multi-hit (eg Jack’s Oligo model, then Moore et al direct inflamm/OPC or via astrocytes)

CNS compartmentalized – evidence late (Renyolds); early (Lucchinetti): microglia – Nat Neurosci

How soon might it start – PD MS evidence (imaging, recent pathology - Bruck)?

Multi-focal (and diffuse) injury evolving over time

January 2002  February 2003  November 2004
Outline

Concepts of inflammation and degeneration in MS
Adaptive and Innate immune responses
Inflammation invoked in Neurodegenerative Diseases
Emerging roles of inflammation impacting:
   (i) Glial influences on neuronal physiology
   (ii) Immune:Glial interactions impacting neurons

Multi-hit model
Direct and Indirect immune effects
How soon might it start?
Degenerative injury
Inflammatory Injury
Imaging Course
Clinical Course
Underlying Biologies
Adaptive
Innate
T cells
B cells
Myeloid
NK cells
Tissue
1. Inflammatory/Degenerative? Different inflammatory ‘flavors’?
2. Relation of Red to Blue? What if we eliminate blue?
3. What starts it all (‘the ‘chicken or the Egg’)?
Inflammation and Degeneration in MS

“An ongoing quandary in MS is what causes the ultimate loss of axons and neurons that underlies disability progression.

Is there an evolving profile of [CNS] inflammation?

Is there another process of neurodegeneration?

If both, are these processes independent or somehow inter-related?”

After Wolfgang Brück

Mechanisms that may underlie Progressive (‘non relapsing’) CNS injury

Inflammation: Perivascular lesion (Adaptive)
Focal and diffuse (Glial activation)
Meningeal inflammation (‘B-cell rich’)

Degeneration: ‘Toxins’: glutamate; O₂, Nitrogen, Fe/Heme
Mitochondrial (demand outstrips supply…)
‘Functional channelopathies’
Neural-glial uncoupling; functional networks

Mechanisms above also likely contribute to limited repair

Loss of compensatory mechanisms + Ageing
Mechanisms that may underlie Progressive (‘non relapsing’) CNS injury

**Inflammation:**
- Adaptive (perivascular lesion?)
- Innate (glial activation; focal + diffuse)
- Immune: glial interactions; meningeal

**Degeneration:**
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Adaptive immune responses in MS

**CNS-antigen directed immune responses**

**T cells:** Th1, Th2, Th9, Th17; Treg…

**B cells:** Plasmablasts/plasma cells, Abs …
- Regulatory (Breg), effector (Beff)

Quite strongly implicated in RRMS
May also be implicated in progressive MS
(Immune cells persisting in CNS? Antibodies?)

Yet: Relevant antigenic-specificities not fully elucidated (multiple reasons)…
Adaptive immune responses in MS

Functional inflammatory profiles distinguish myelin-reactive T cells from patients with multiple sclerosis

Yonghao Cao,1* Brittany A. Goods,2* Khadir Raddassi,1 Gerald T. Nepom,3 William W. Kwok,3,4 J. Christopher Love,5,6,7 David A. Hafler1


Abnormal responses of myelin-reactive T cells in MS

Mechanisms that may underlie Progressive ('non relapsing’) CNS injury

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**Innate inflammation in multiple neurodegenerative diseases**


**Chronic Epilepsy:** NLRP1 inflammasome is activated in patients with TLE and contributes to neuronal pyroptosis. Tan CC, et al. J Neuroinflammation. 2015


Are there common mechanisms across diseases?
Comparative genome-wide expression data (Illumina H-Ref 8) AzD, ALS, HD, MS, PD, MS and Schizophrenia (n = 113 well-characterized post-mortem brain tissues).
Results: no dysregulated gene (passing QC) found across all 61 dysregulated genes shared when comparing 4+5 diseases)
Hints for common neuronal homeostatic, survival and synaptic plasticity pathways.
All diseases exhibited changes in several inflammation-related genes …role of the innate immune system in the pathogenesis and/or response to neurodegeneration.

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Concepts of inflammation and degeneration in MS
Inflammation invoked in Neurodegenerative Diseases
Emerging roles of:
(i) Glial influences on neuronal physiology
(ii) Immune:Glial interactions impacting neurons
Cellular and molecular mechanisms by which inflammation may contribute to CNS degeneration
How soon might it start?
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Glial cells in progressive CNS injury and repair

Astrocyte and Microglia Biology: Novel functions and distinct subsets impacting OPC and Neurons

Glia-glia interactions: astrocyte and microglia effects on OPC during development and demyelinating disease.

Astrocytes-neuron signaling in brain.

Molecular and functional microglia signatures
Several important neuronal–glial interactions recognized

Synapse loss is striking in neurodegenerative disease and Glia have intimate roles in synapse physiology:

Astrocytes, OL, and microglia: All crucial and multifaceted roles in maintenance of synaptic function and excitability.

Neuroinflammation contribution to synapse loss may be primarily mediated by altered glial functions.

Question: how does Inflammation (aging, neuronal stress) impact on glia (eg activation) → neurons/synapses

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**Microglia:** Inflammation → glial response → Neurodegeneration


System xC- is a mediator of microglial function; its deletion slows symptoms in ALS mice. Mesci P, et al Brain. 2015.


Possible Mechanisms of Inflammation → Degeneration

Adaptive Immune:

Innate immune: Inflammasome, TLR, Complement, Vesicles:


Neuroinflammation: Take the Bad with the Good?


Microglia signature (gene expression and quantitative mass spectrometry) suggests ‘Maintainers of CNS quiescence’


However:

TREM2 Lipid Sensing Sustains the Microglial desired Response in an Alzheimer's Disease Model.
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‘Multi-hit’ Model of Oligodendrocyte Injury

- Inflammation
- Ischemia
- Infection
- Trauma

p53 upregulation

IFNg
TNFa

ROS

Death receptor upregulation
Fas
DR4/5

Death receptor triggering and apoptotic signalling

IFNg
TNFa

T cell

Microglia

ROS

From Antel J. Clin Neurol Neurosurg. 2005
‘Multi-hit’ Model of Oligodendrocyte Injury

- Inflammation
- Ischemia
- Infection
- Trauma

p53 upregulation

Death receptor upregulation

Sublethal injury

Death receptor triggering and apoptotic signalling

Lethal injury

From Antel J. Clin Neurol Neurosurg. 2005

Human neurons live on ‘bed’ of astrocytes

Immune cells can injure astrocytes with secondary injury to neurons


Direct and Indirect Effects of Immune and Central Nervous System–Resident Cells on Human Oligodendrocyte Progenitor Cell Differentiation

Craig S. Moore, Qiao-Ling Cui, Nebras M. Warsi, Bryce A. Durafourt, Nika Zorko, David R. Owen, Jack P. Antel, and Amit Bar-Or

The Journal of Immunology, 2015, 194: 761–772.
Meningeal inflammation in MS can be ‘B cell rich’


Howell OW, et al Brain. 2011
Cortical lesions: correlation with disease course, including disability and cognitive deficits (Howell OW, et al Brain. 2011 Sep;134(Pt 9):2755-71)


Human Glial Cells/Neuroglia

- Trypsin
- DNase
- Wash
- Percoll to remove myelin
- 5% MEM/DMEM
- Microglia adherent
- Oligos, others float

Microglia
Astrocyte
Oligodendrocyte
OPC
Neurite outgrowth is differentially impacted by distinct immune cell subsets
Madeline Pool a, Isabel Rambaldi a, Peter J. Darlington a,b, Melissa C. Wright a,b, Alyson E. Fournier a,b, Amit Bar-Or a,b,++

Short communication
Myeloid lineage cells inhibit neurite outgrowth through a myosin II-dependent mechanism
Madeline Pool a, Isabel Rambaldi a, Bryce A. Duraufort b, Melissa C. Wright a,b, Jack P. Antel b, Amit Bar-Or a,b,+,1, Alyson E. Fournier a,b,+,1
‘State’ of myeloid cell: Balance between Activation and Quiescence/Inhibitory molecules

Adapted from Kierdorf et al., Frontiers in Cell Neuroscience 2013

Methods: effects of Breg and Beff products on microglia and macrophage Activation/Quiescence molecules

Hanane Touil
Effects of Breg & Beff products on activation molecule expression by human macrophage and microglia

Human Macrophages

- CD80
- CCR7
- HLA-DR

Human Microglia

- CD80
- CCR7
- HLA-DR

Effects of Breg & Beff products on quiescence/inhibitory molecule expression by human macrophage and microglia

Human Macrophages

- TREM-2
- SIRP1α
- mCsfr

Human Microglia

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Summary: Inflammation in Progressive MS

Inflammation appears to be involved throughout MS spectrum; contributes in different ways in different CNS sub-compartments

- Perivascular, diffuse, meningeal
- Roles for both adaptive and innate; likely Innate > Adaptive
- Both direct and indirect injury mechanisms; 'multiple hit' model
- Glial cells (microglia, astrocytes) as mediators of inflammation
- Important interface between inflammatory and degenerative mechanisms

‘Progressive disease biology’ may start earlier than we would like to think!
Summary: Inflammation and Neurodegeneration

Valuable insights from comparing across diseases (Similarities and important differences)

Emerging roles of glial cells: neuronal integrity/function

Underscores importance of understanding both Neuronal:glial and Glial:Glial interactions

Neuroinflammation \(\rightarrow\) Glial state \(\rightarrow\) Neurodegeneration

Mechanisms may be starting at the very beginning…
Two Views on Neuro-Immunology of MS

A. Dysregulated Immune Response → Inflammatory CNS Injury → Degenerative CNS Injury

B. Dysregulated Immune Response → Degenerative CNS Injury → Inflammatory CNS Injury

Bar-Or A. Advances in Neurology; 23:149-175, 2006
Pediatric MS CSF may better reflect Early MS targets

**Hypothesis**: Compact myelin antigens (MBP, PLP), traditionally considered disease-initiating, will be over-represented in CSF of children with MS than controls

**Approach**: Blinded ‘unbiased’ proteomics of CSF obtained at time of initial episode of pediatric CNS inflammatory demyelination*

Subsequent ascertainment as MS (vs monophasic)

* Canadian Pediatric Demyelinating Disease Study

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**Implication of Perturbed Axoglial Apparatus in Early Pediatric Multiple Sclerosis**

Ajit Singh Dhaunchak, PhD,1,2 Christopher Becker, PhD, Howard Schulman, PhD, Omar De Faria, Jr, MSc, Sathyannath Rajasekhara, PhD, Brenda Banwell, MD, David R. Colman, PhD, and Amit Bar-Or, MD, on behalf of the Canadian Pediatric Demyelinating Disease Group

ANN NEUROL 2012;71:601–613
Less known features:

? Impact of ‘peripheral/relapsing inflammation’ on ‘CNS-compartmentalized inflammation’

Eg: peripheral immune cell responses impacting:
- microglial activation
- glia-glia interactions

? Impact of ‘CNS inflammation’ (whether peripheral or compartmentalized) on ‘non-inflammatory’ (‘degen’) aspects of disease:
- inflammation impacting mitochondrial function
- Klaus Nave presentation at ISNI/Mahad

axon loss in progressive MS: three step hypothesis

1. formation
2. amplification
3. aberrant placement

- Mahad, Trapp & Lassmann, Lancet Neurol 2014
- Campbell, Worrall & Mahad, MSJ 2014
- Ohno et al, PNAS 2014
Meningeal inflammation in MS is ‘B cell rich’ *

* Staining with aCD20 (B cell marker)

Secretory products of multiple sclerosis B cells are cytotoxic to oligodendroglia in vitro

Robert P. Lisak a,b,*, Joyce A. Benjamins a,b, Liljana Nedelkoska a, Jennifer L. Barger a, Samia Ragheb a,b, Boli Fan c,d, Nadia Ouamara c,d, Trina A. Johnson c,d, Sathyath Rajasekharan c,d, Amit Bar-Or c,d,e,f,*

Journal of Neuroimmunology 246 (2012) 85-95
Biologies Contributing to Central Nervous System Injury in MS


RMS = relapsing multiple sclerosis; SPMS = secondary progressive MS; PPMS = primary progressive MS.

Identification of a microglia signature by gene expression and quantitative mass spectrometry (AffyExon1 & MG400 chip)

Identification of a unique TGF-β-dependent molecular and functional signature in microglia

NATURE NEUROSCIENCE VOLUME 17 | NUMBER 1 | JANUARY 2014

Identification of a microglia signature by gene expression and quantitative mass spectrometry (AffyExon1 & MG400 chip)

Immune ablation and autologous stem cell reconstitution* (BMT) essentially halts measures of new focal disease activity

* With M. Freedman, H. Atkins, D. Arnold, J. Chen, and the Canadian Collaborative BMT in MS Study Group

Darlington et al, Ann Neurol, 2013

Effects of B cell Depletion in PPMS

Effects of B cell Depletion in PPMS

Preplanned subgroup analyses

Presence of IgM OCB (M+) was associated strongly with presence of Gd+ lesions in PPMS

Gd+ lesions continued in M+ treated with placebo..
No new Gd+ lesions in M+ treated with rituximab.
B cell depletion decreased IgM OCB but not IgG OCB:
Secretory products of multiple sclerosis B cells are cytotoxic to oligodendroglia in vitro

Robert P. Lisak a,b,c, Joyce A. Benjamins a,e, Liliana Nedelkoska a, Jennifer L. Barger a, Samia Ragheb a,b, Boli Fan c, Nadia Ouamara a,c, Trina A. Johnson c,d, Sathyarnath Rajasekharan e,c,f, Amit Bar-Or c,d,e,f,g

Journal of Neuroimmunology 246 (2012) 85–95

Immunoglobulin M Oligoclonal Bands: Biomarker of Targetable Inflammation in Primary Progressive Multiple Sclerosis

Luisa M. Villar, PhD,¹ Bonaventura Casanova, MD,⁎ Nadia Ouamara, MSc,³
Manuel Comabella, MD,¹ Farzaneh Jalili,⁵ David Leppert, MD,⁶
Clara de Andrés, MD,⁷ Guillermo Izquierdo, MD,⁸ Rafael Arroyo, MD,⁹
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Evgeniy Evdoshenko, MD,¹⁵ José C. Álvarez-Cermeño, MD,¹,¹⁶ and
Amit Bar-Or. MD. FRCPC,³,⁵

ANN NEUROL 2014;00:000–000
Peripheral Inflammation in PPMS (vs RRMS, SPMS)

Several studies – no clear convergence/conclusion


“We found unique autoantibody patterns that distinguished RRMS, secondary progressive (SPMS), and primary progressive (PPMS) MS from both healthy controls and other neurologic or autoimmune driven diseases including Alzheimer's disease, adrenoleukodystrophy, and lupus erythematosus.”

Lack of Replication; technology platforms; Cohort matching; (eg disease duration; Immune senescence)

Not convinced that relevant differences have been established between peripheral immune responses in PPMS and SPMS